

FIGURE 1

APGK12 (SEQ ID NO 1) Replicon RNA compared to I377/NS2-3' Replicon RNA

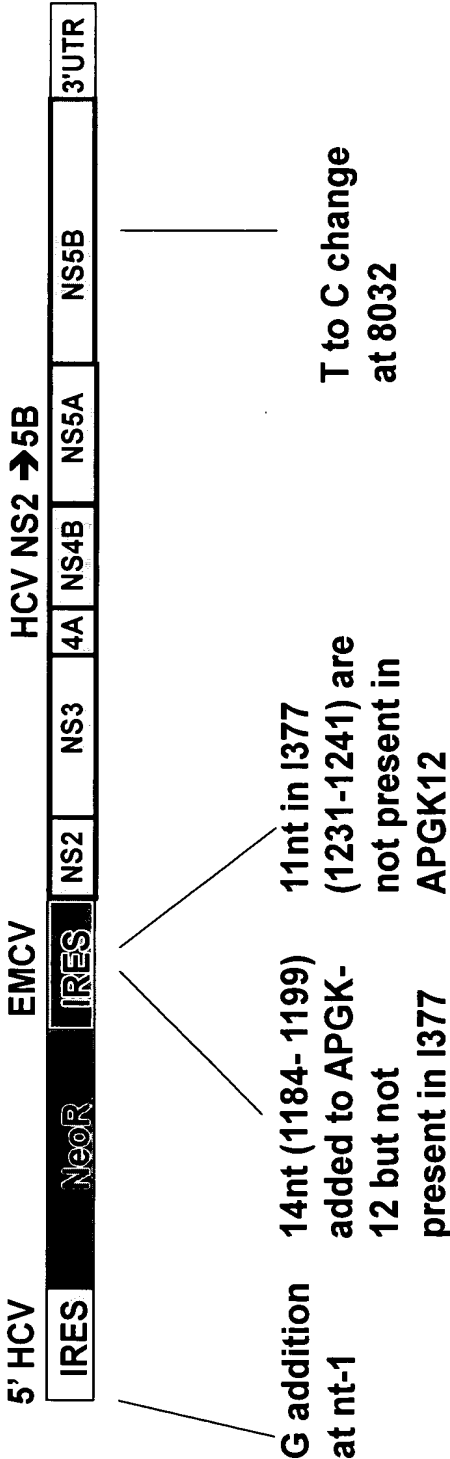


FIGURE 2

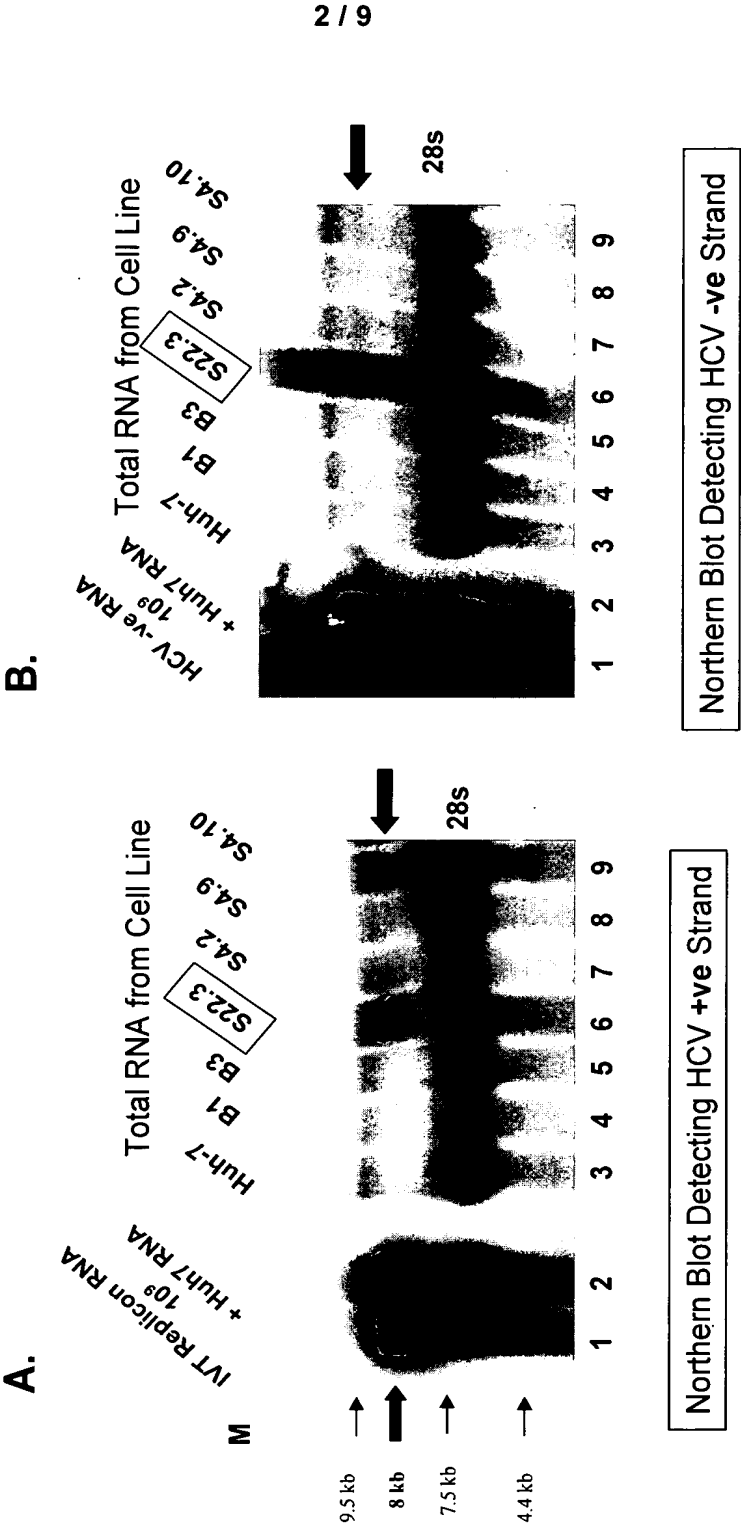


FIGURE 3

A.
S22.3

B.

**Anti-HCV NS4A
Immunofluorescence**

DIC Microscopy

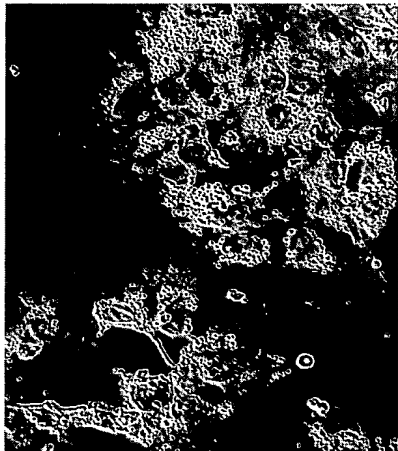
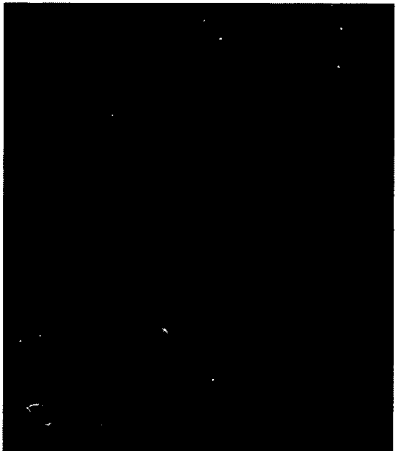


FIGURE 4

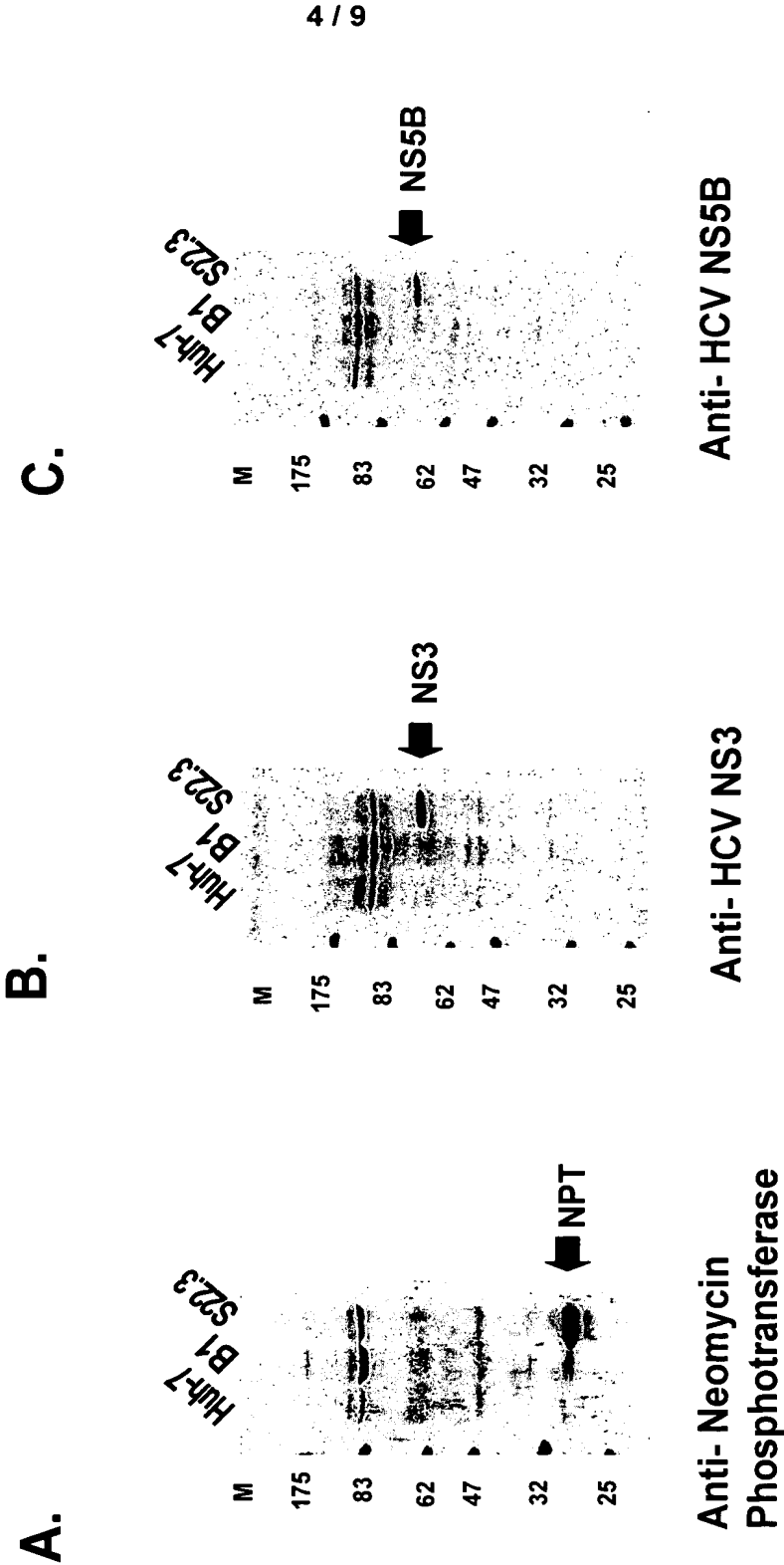


FIGURE 5A

	S 22-3 SEQ ID NO. 2	R3 SEQ ID NO. 4	R3-rep SEQ ID NO. 7	R7 SEQ ID NO. 5	R16 SEQ ID NO. 6
5'end - FIRST nt (HCV IRES)	*G (nt 1) A	G (nt 1) A	-	-	G (nt 1) A
Neo	-	A (nt 461) G	-	-	-
EMCV IRES	-	A (nt 1739) G	-	-	-
NS 2	-	-	-	-	-
NS 3	-	G (nt 2778) A A (nt 2840) C A (nt 4052) G	T (nt 2509) C G (nt 2778) A A (nt 2840) C T (nt 3574) C A (nt 4052) G	A (nt 2935) G A (nt 2979) G	A (nt 2816) G A (nt 2979) G
NS 4A	A (nt 4446) R	A (nt 4446) G	C (nt 4387) T A (nt 4446) G C (nt 4507) T	-	C (nt 4475) T
NS 4B	-	T (nt 4855) C	T (nt 4855) C	-	-
NS 5A	G (nt 5498) T A (nt 6288) R	A (nt 5351) G G (nt 5498) T G (nt 5659) A C (nt 5871) T A (nt 6288) G	A (nt 5351) G G (nt 5498) T G (nt 5659) A T (nt 5838) C C (nt 5871) T A (nt 6115) G	A (nt 5324) G G (nt 5498) T T (nt 6001) C	G (nt 5498) C T (nt 6320) C T (nt 6584) C
NS 5B	-	A (nt 6652) G	-	C (nt 7252) T T (nt 8349) C	-
3'end - last 98 nt	-	-	-	-	-

*first nt = G from HCV IRES

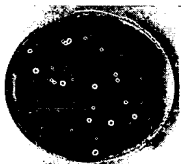
FIGURE 5B

	S 22-3 SEQ ID NO. 2	R3 SEQ ID NO. 4	R3 Rep SEQ ID NO. 7	R7 SEQ ID NO. 5	R16 SEQ ID NO. 6
	G (nt 1) A	G (nt 1) A	-	-	G (nt 1) A
5'end - FIRST nt (HCV IRES)					
NS 2	-	-	-	-	-
NS 3	-	R (1135) K S (1560) G	R (1135) K S (1560) G	E (1202) G	S (1148) G E (1202) G
NS 4A	K (1691) mix K/R	K (1691) R	K (1691) R	-	L (1701) F
NS 4B	-	-	-	-	-
NS 5A	G (2042) C	T (1993) A G (2042) C P (2166) L	T (1993) A G (2042) C L (2155) P P (2166) L	I (1984) V G (2042) C	G (2042) R S (2404) P
NS 5B	-	-	-	M (2992) T	-
3'end - last 98 nt	-	-	-	-	-

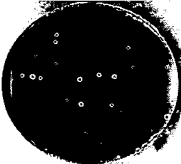
first a.a. of NS2 = 810

FIGURE 6
AMINO ACID SUBSTITUTIONS

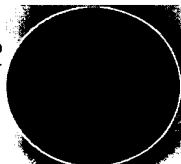
CLONE APGK-12		EMCV		HCV NS2→5B				3'HCV		
5' HCV	Neor	IRES	NS2	NS3	4A	NS4B	NS5A	NS5B	UTR	
G (nt1) SEQ ID NO 1										
A (nt1) SEQ ID NO 24			-	-	-	-	-	-		
R3 rep A(nt1) SEQ ID NO 25				R(1135)K S(1560)G	K(1691)R	-	T(1993)A G(2042)C L(2155)P P(2166)L			
G(nt1) SEQ ID NO 7				R(1135)K S(1560)G	K(1691)R	-	T(1993)A G(2042)C L(2155)P P(2166)L			



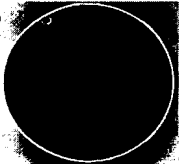
77 cfu/μg



86 cfu/μg



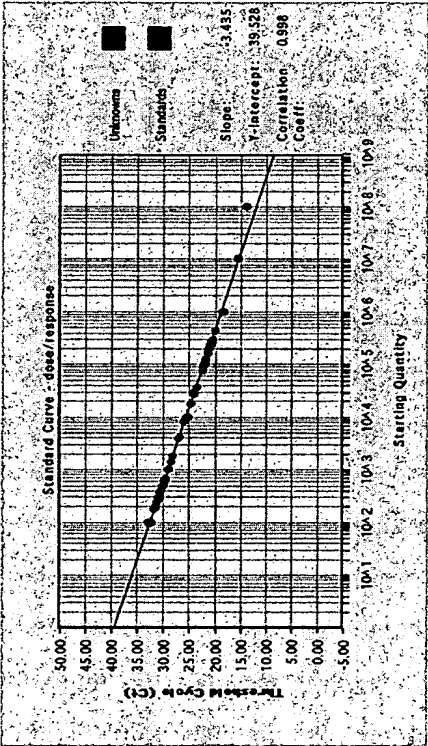
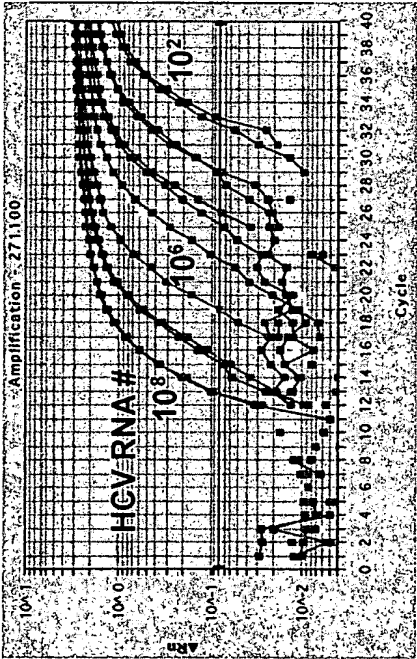
1100000cfu/μg



2000000cfu/μg

FIGURE 7

HCV-Replicon: RNA Quantification



Ct = Threshold cycle \propto Starting RNA Quantity

FIGURE 8

